

Package ‘SimFFPE’

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Type Package

Title NGS Read Simulator for FFPE Tissue

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Description The NGS (Next-Generation Sequencing) reads from FFPE (Formalin-Fixed Paraffin-Embedded) samples contain numerous artifact chimeric reads (ACRS), which can lead to false positive structural variant calls. These ACRs are derived from the combination of two single-stranded DNA (ss-DNA) fragments with short reverse complementary regions (SRCRs). This package simulates these artifact chimeric reads as well as normal reads for FFPE samples on the whole genome / several chromosomes / large regions.

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SimFFPE-package	<i>NGS Read Simulator for FFPE Tissue</i>
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Description

The NGS (Next-Generation Sequencing) reads from FFPE (Formalin-Fixed Paraffin-Embedded) samples contain numerous artifact chimeric reads (ACRS), which can lead to false positive structural variant calls. These ACRs are derived from the combination of two single-stranded DNA (ss-DNA) fragments with short reverse complementary regions (SRCRs). This package simulates these artifact chimeric reads as well as normal reads for FFPE samples on the whole genome / several chromosomes / large regions.

Details

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The NGS (Next-Generation Sequencing) reads from FFPE (Formalin-Fixed Paraffin-Embedded) samples contain numerous artifact chimeric reads (ACRs), which can lead to false positive structural variant calls. These ACRs are derived from the combination of two single-stranded DNA (ss-DNA) fragments with short reverse complementary regions (SRCR). This package simulates these artifact chimeric reads as well as normal reads for FFPE samples. To simplify the simulation, the genome is divided into small windows, and SRCRs are found within the same window (adjacent ss-DNA combination) or between different windows (distant ss-DNA simulation). For adjacent ss-DNA combination events, the original genomic distance between and strands of two combined SRCRs are also simulated based on real data. The simulation can cover whole genome, or several chromosomes, or large regions, or whole exome, or targeted regions. It also supports enzymatic / random fragmentation and paired-end / single-end sequencing simulations. Fine-tuning can be achieved by adjusting the parameters, and multi-threading is supported. Please check the package vignette for the guidance of fine-tuning
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There are three available functions for NGS read simulation of FFPE samples:

1. `calcPhredScoreProfile`: Calculate positional Phred score profile from BAM file for read simulation.
2. `readSimFFPE`: Simulate artifact chimeric reads on whole genome, or several chromosomes, or large regions.
3. `targetReadSimFFPE`: Simulate artifact chimeric reads in exonic / targeted regions.

Author(s)

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See Also

[calcPhredScoreProfile](#), [readSimFFPE](#), [targetReadSimFFPE](#)

Examples

```
PhredScoreProfilePath <- system.file("extdata", "PhredScoreProfile2.txt",
                                     package = "SimFFPE")
PhredScoreProfile <- as.matrix(read.table(PhredScoreProfilePath, skip = 1))
colnames(PhredScoreProfile) <-
  strsplit(readLines(PhredScoreProfilePath)[1], "\t")[[1]]

referencePath <- system.file("extdata", "example.fasta", package = "SimFFPE")
reference <- readDNAStringSet(referencePath)

## Simulate reads of the first three sequences of the reference genome

sourceSeq <- reference[1:3]
outFile1 <- paste0(tempdir(), "/sim1")
readSimFFPE(sourceSeq, referencePath, PhredScoreProfile, outFile1,
            coverage = 80, enzymeCut = TRUE, threads = 2)

## Simulate reads for targeted regions

bamFilePath <- system.file("extdata", "example.bam", package = "SimFFPE")
regionPath <- system.file("extdata", "regionsBam.txt", package = "SimFFPE")
regions <- read.table(regionPath)
PhredScoreProfile <- calcPhredScoreProfile(bamFilePath, targetRegions = regions)

regionPath <- system.file("extdata", "regionsSim.txt", package = "SimFFPE")
targetRegions <- read.table(regionPath)

outFile <- paste0(tempdir(), "/sim2")
targetReadSimFFPE(referencePath, PhredScoreProfile, targetRegions, outFile,
                  coverage = 80, readLen = 100, meanInsertLen = 180,
                  sdInsertLen = 50, enzymeCut = FALSE)
```

calcPhredScoreProfile *Estimate Phred score profile for FFPE read simulation*

Description

Calculate Phred score profile from the entire BAM file or reads in subsampled regions.

Usage

```
calcPhredScoreProfile(bamFilePath, mapqFilter = 0, maxFileSize = 1,
  targetRegions = NULL, subsampleRatio = NA, subsampleRegionLength = 1e+05,
  disableSubsampling = FALSE, threads = 1)
```

Arguments

<code>bamFilePath</code>	BAM file to be processed.
<code>mapqFilter</code>	Filter for mapping quality. Reads with mapping quality below this value will be excluded from calculation.
<code>maxFileSize</code>	The maximum file size (in GB) that allows processing of the entire BAM file. If <code>disableSubsampling</code> is set to false, BAM file larger than this size will be subsampled for calculation.
<code>targetRegions</code>	A <code>DataFrame</code> or <code>GenomicRanges</code> object representing target regions for calculation. Use it for targeted sequencing / WES data, or when you need to manually select subsampled regions (set <code>disableSubsampling</code> to true in this case). If it is a <code>DataFrame</code> , the first column should be the chromosome, the second the start position and the third the end position. Please use one-based coordinate systems (the first base should be marked with 1 but not 0).
<code>subsampleRatio</code>	Subsample ratio. Together with <code>subsampleRegionLength</code> to determine subsampled regions. When <code>subsampleRatio</code> is not given, it will be assigned the value of <code>maxFileSize</code> divided by the input BAM file size. Range: 0 to 1.
<code>subsampleRegionLength</code>	Length of each subsampled region. Unit: base pair (bp).
<code>disableSubsampling</code>	Force to use the entire BAM file for calculation when set to true.
<code>threads</code>	Number of threads used. Multi-threading can speed up the process.

Details

Calculate positional Phred score profile from the entire BAM file or reads in subsampled regions. A Phred score profile will be returned, which can then be used in read simulation.

Value

A matrix will be returned. Each row of the matrix represents a position in the read (from begin to end), and each column the Phred quality score of base-calling error probabilities. The value in the matrix represents the positional Phred score proportion.

Author(s)

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See Also

[SimFFPE](#), [readSimFFPE](#), [targetReadSimFFPE](#)

Examples

```
bamFilePath <- system.file("extdata", "example.bam", package = "SimFFPE")
regionPath <- system.file("extdata", "regionsBam.txt", package = "SimFFPE")
regions <- read.table(regionPath)
PhredScoreProfile <- calcPhredScoreProfile(bamFilePath, targetRegions = regions)
```

readSimFFPE	<i>Simulate normal and artifact chimeric reads in NGS data of FFPE samples for whole genome / several chromosomes / large regions</i>
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Description

NGS data from FFPE samples contain numerous artifact chimeric reads. These chimeric reads are formed through the combination of two single-stranded DNA (ss-DNA) with short reverse complementary regions (SRCR). This function simulates these artifact chimeric reads as well as normal reads for FFPE samples on the whole genome, or several chromosomes, or large regions. To simplify the simulation, the genome is divided into small windows, and SRCRs are found within the same window (adjacent ss-DNA combination) or between different windows (distant ss-DNA simulation).

Usage

```
readSimFFPE(sourceSeq, referencePath, PhredScoreProfile, outFile, coverage,
readLen=150, meanInsertLen=250, sdInsertLen=80, enzymeCut=FALSE,
chimericProp=0.1, sameChrProp=0.43, windowLen=5000, adjChimProp=0.63,
sameStrandProp=0.65, meanLogSRCRLen=1.8, sdLogSRCRLen=0.55, maxSRCRLen=32,
meanLogSRCRDist=4.7, sdLogSRCRDist=0.35, distWinLen=5000, spikeWidth = 1500,
betaShape1=0.5, betaShape2=0.5, sameTarRegionProb=0, adjFactor=1.65,
distFactor=1.65, chimMutRate=0.003, noiseRate=0.0015, highNoiseRate=0.08,
highNoiseProp=0.01, pairedEnd=TRUE, prefix="SimFFPE", threads=1,
adjChimeric=TRUE, distChimeric=TRUE, normalReads=TRUE, overWrite=FALSE)
```

Arguments

sourceSeq	A DNASTringSet object of DNA sequences used for simulation. It can cover the entire reference genome or selected chromosomes or chromosome regions.
referencePath	Path to the reference genome.
PhredScoreProfile	A matrix representing the positional Phred score proportion. Each row of the matrix represents a position in the read (from begin to end), and each column the Phred quality score of base-calling error probabilities. The profile can be calculated from BAM file using the calcPhredScoreProfile function.
outFile	Output file path for the FASTQ file with simulated reads. Please include the name of the output file without extension, e.g. "/tmp/sim1".
coverage	Coverage of the simulation.
readLen	Read length of the simulation.

meanInsertLen	Mean insert length for the simulation (normally distributed).
sdInsertLen	Standard deviation of the insert length for simulation (normally distributed).
enzymeCut	Simulate enzymatic fragmentation if it is set to true, otherwise simulate random fragmentation.
chimericProp	Proportion of artifact chimeric fragments (chimeric fragments / chimeric or normal fragments). Range: 0 to 1.
sameChrProp	Proportion of artifact chimeric fragments that are derived from the combination of two ss-DNA coming from the same chromosome. Range: 0 to 1.
windowLen	The window length used in adjacent ss-DNA combination simulation. To simulate adjacent ss-DNA combinations, input DNA sequences are divided into small windows of equal size, and short reverse complementary regions are searched within the same window to form artifact chimeric fragments. Unit: base pair (bp).
adjChimProp	Proportion of adjacent ss-DNA combinations among same chromosomal ss-DNA combinations. Range: 0 to 1.
sameStrandProp	Proportion of same-strand ss-DNA combinations among adjacent ss-DNA combinations. For paired end sequencing, the larger the proportion, the greater the proportion of improperly paired reads with LL / RR pair orientation, and the smaller the proportion with RL pair orientation. Range: 0 to 1.
meanLogSRCRLen	Mean of log scaled length of the short reverse complementary regions (SRCR) in artifact chimeric fragments. SRCRs links two ss-DNA together, yielding artifact chimeric fragments. The length of SRCR follows a log-normal distribution. See rlnorm for more details. Unit: base pair (bp).
sdLogSRCRLen	Standard deviation of log scaled length of the short reverse complementary regions.
maxSRCRLen	Maximum length of the short reverse complementary regions. Unit: base pair (bp).
meanLogSRCRDist	Mean of log scaled original genomic distance of the short reverse complementary regions(SRCR) in artifact chimeric fragments. SRCRs links two ss-DNA together, yielding artifact chimeric fragments. The distance of SRCR is the original genomic distance between the two short reverse complementary segments, which follows a log-normal distribution in simulation. For log-normal distribution, see rlnorm for more details. Unit: base pair (bp).
sdLogSRCRDist	Standard deviation of log scaled original genomic distance of the short reverse complementary regions(SRCR) in artifact chimeric fragments.
distWinLen	The window length used in distant ss-DNA simulation. To simulate distant ss-DNA combinations, the short reverse complementary regions(SRCR) are searched between different windows. Unit: base pair (bp).
spikeWidth	The width of chimeric read spike used to simulate distant ss-DNA combinations. In real FFPE samples, the chimeric reads formed by distant DNA combination are unevenly distributed along the chromosome. Some regions are enriched in these reads while some others are scarce. The length of these regions are of similar scale; therefore, a defined width is used for simulation. Suggested range: 1500-2000. Unit: base pair (bp).

betaShape1	Shape parameter a of beta distribution used to model the unevenly distributed distant ss-DNA combinations. The number of seeds in each "spike" follows a "U" shaped beta distribution. Use this parameter to adjust the shape of the curve. See rbeta for more details. Range: 0-1.
betaShape2	Shape parameter b of beta distribution used to model the unevenly distributed distant ss-DNA combinations. The number of seeds in each "spike" follows a "U" shaped beta distribution. Use this parameter to adjust the shape of the curve. See rbeta for more details. Range: 0-1.
sameTarRegionProb	Probability of two distant ss-DNA combination events coming from the same two different windows.
adjFactor	Increase this value if the number of simulated adjacent chimeric reads is smaller than expected ($\text{sameChrProp} * \text{adjChimProp}$), decrease if the opposite is true.
distFactor	Increase this value if the number of simulated distant chimeric reads is smaller than expected, decrease if the opposite is true.
chimMutRate	Mutation rate for each base in chimeric fragments. In the chimeric fragment formation process, biological-level errors might occur and lead to mutations on these artifact fragments. For all four basic types of nucleotides, the substitution probability is set equal. Range: 0-0.75.
noiseRate	Noise rate for each base in reads. This is used for sequencing-level errors. The probability is set equal for all four basic types of nucleotides. Range: 0-0.75.
highNoiseRate	A second noise rate for each base in reads. In some real sequencing data, some reads are much more noisy than others. This parameter can be used for this situation. Range: 0-0.75.
highNoiseProp	Proportion of reads to be simulated with highNoiseRate other than noiseRate. Range: 0-1.
pairedEnd	Simulate paired end sequencing when set to true.
prefix	Prefix for read names. When reads from different runs of simulation have to be merged, please make sure that they have different prefixes.
threads	Number of threads used. Multi-threading can speed up the process.
adjChimeric	Generate reads from adjacent ss-DNA combinations if it is set to true. If it is set to false, skip this process.
distChimeric	Generate reads from distant ss-DNA combinations if it is set to true. If it is set to false, skip this process.
normalReads	Generate reads from normal fragments if it is set to true. If it is set to false, skip this process.
overWrite	Overwrite the file if file with the same output path exists and it is set to true. If file with same output path exists and it is set to false, reads will be appended to the existing file.

Details

The NGS (Next-Generation Sequencing) reads from FFPE (Formalin-Fixed Paraffin-Embedded) samples contain numerous artifact chimeric reads (ACRS), which can lead to false positive structural variant calls. These ACRs are derived from the combination of two single-stranded DNA (ss-DNA) fragments with short reverse complementary regions (SRCR). This function simulates these

artifact chimeric reads as well as normal reads for FFPE samples on the whole genome / several chromosomes / large regions. To simplify the simulation, the genome is divided into small windows, and SRCRs are found within the same window (adjacent ss-DNA combination) or between different windows (distant ss-DNA simulation). For adjacent ss-DNA combination events, the original genomic distance between and strands of two combined SRCRs are also simulated based on real data. In the output fastq file, reads are distinguished by prefixes "adjChimeric", "distChimeric" and "Normal" in their names. The parameter PhredScoreProfile can be calculated by the function [calcPhredScoreProfile](#). To simulate whole exome sequencing (WES) or targeted sequencing, please use the function [targetReadSimFFPE](#).

Value

NULL. Reads will be written to the output FASTQ file.

Note

When fine-tuning is needed, simulate reads from certain areas / chromosomes instead of the entire genome to save the run-time. Please check the package vignette for the guidance of fine-tuning.

Author(s)

Lanying Wei <lanying.wei@uni-muenster.de>

See Also

[SimFFPE](#), [calcPhredScoreProfile](#), [targetReadSimFFPE](#)

Examples

```
PhredScoreProfilePath <- system.file("extdata", "PhredScoreProfile2.txt",
                                     package = "SimFFPE")
PhredScoreProfile <- as.matrix(read.table(PhredScoreProfilePath, skip = 1))
colnames(PhredScoreProfile) <-
  strsplit(readLines(PhredScoreProfilePath)[1], "\t")[[1]]

referencePath <- system.file("extdata", "example.fasta", package = "SimFFPE")
reference <- readDNASTringSet(referencePath)

## Simulate reads of the first three sequences of reference genome

sourceSeq <- reference[1:3]
outFile1 <- paste0(tempdir(), "/sim1")
readSimFFPE(sourceSeq, referencePath, PhredScoreProfile, outFile1,
            enzymeCut = FALSE, coverage=80, threads = 2)

## Simulate reads of defined regions on the first two sequences of reference
## genome

sourceSeq2 <- DNASTringSet(lapply(reference[1:2], function(x) x[1:10000]))
outFile2 <- paste0(tempdir(), "/sim2")
```



```

readSimFFPE(sourceSeq2, referencePath, PhredScoreProfile, outFile2,
             coverage = 80, enzymeCut = TRUE, threads = 1)

## Simulate reads of defined regions on the second and the third sequence of
## reference genome and merge them with existing reads (a different prefix is
## needed)

sourceSeq3 <- DNASTringSet(lapply(reference[2:3], function(x) x[1:10000]))
readSimFFPE(sourceSeq3, referencePath, PhredScoreProfile, outFile2,
             prefix = "simFFPE2", coverage = 80, enzymeCut = TRUE,
             threads = 1, overWrite = FALSE)

```

targetReadSimFFPE	<i>Simulate normal and artifact chimeric reads in NGS data of FFPE samples for exonic / targeted regions</i>
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Description

NGS data from FFPE samples contain numerous artifact chimeric reads. These chimeric reads are formed through the combination of two single-stranded DNA (ss-DNA) with short reverse complementary regions (SRCR). This function simulates these artifact chimeric reads as well as normal reads for FFPE samples within defined regions. To simplify the simulation, the genome is divided into small windows, and SRCRs are found within the same window (adjacent ss-DNA combination) or between different windows (distant ss-DNA simulation).

Usage

```

targetReadSimFFPE(referencePath, PhredScoreProfile, targetRegions, outFile,
                  coverage, readLen=150, meanInsertLen=250, sdInsertLen=80, enzymeCut=FALSE,
                  padding=50, minGap=5, chimericProp=0.1, sameChrProp=0.43, windowLen=5000,
                  adjChimProp=0.63, sameStrandProp=0.65, meanLogSRCRLen=1.8, sdLogSRCRLen=0.55,
                  maxSRCRLen=32, meanLogSRCRDist=4.7, sdLogSRCRDist=0.35, distWinLen=5000,
                  spikeWidth=1500, betaShape1=0.5, betaShape2=0.5, sameTarRegionProb=0,
                  adjFactor = 1.3, distFactor = 1.3, chimMutRate=0.003, noiseRate=0.0015,
                  highNoiseRate=0.08, highNoiseProp=0.01, pairedEnd=TRUE, prefix="SimFFPE",
                  threads=1, adjChimeric=TRUE, distChimeric=TRUE, normalReads=TRUE,
                  overWrite=FALSE)

```

Arguments

referencePath Path to the reference genome.

PhredScoreProfile

A matrix representing the positional Phred score proportion. Each row of the matrix represents a position in the read (from begin to end), and each column the Phred quality score of base-calling error probabilities. The profile can be calculated from BAM file using the [calcPhredScoreProfile](#) function.

targetRegions	A DataFrame or GenomicRanges object representing the exonic / targeted regions to simulate. If it is a DataFrame, the first column should be the chromosome, the second the start position and the third the end position. Please use one-based coordinate systems (the first base should be marked with 1 but not 0).
outFile	Output file path for the FASTQ file with simulated reads. Please include the name of the output file without extension, e.g. "/tmp/sim1".
coverage	Coverage of the simulation.
readLen	Read length of the simulation.
meanInsertLen	Mean insert length for the simulation (normally distributed).
sdInsertLen	Standard deviation of the insert length for simulation (normally distributed).
enzymeCut	Simulate enzymatic fragmentation if it is set to true, otherwise simulate random fragmentation.
padding	Length of padding of input target regions. The padding length will be added to both sides of target regions. Range: natural numbers. Unit: base pair (bp).
minGap	Minimal allowed length of gap between target regions. Regions with a gap smaller than this value will be merged. If this value is not given, the value of input readLen will be used. Range: natural numbers. Unit: base pair (bp).
chimericProp	Proportion of artifact chimeric fragments (chimeric fragments / chimeric or normal fragments). Range: 0 to 1.
sameChrProp	Proportion of artifact chimeric fragments that are derived from the combination of two ss-DNA coming from the same chromosome. Range: 0 to 1.
windowLen	The window length used in adjacent ss-DNA combination simulation. To simulate adjacent ss-DNA combinations, input DNA sequences are divided into small windows of equal size, and short reverse complementary regions are searched within the same window to form artifact chimeric fragments. Unit: base pair (bp).
adjChimProp	Proportion of adjacent ss-DNA combinations among same chromosomal ss-DNA combinations. Range: 0 to 1.
sameStrandProp	Proportion of same-strand ss-DNA combinations among adjacent ss-DNA combinations. For paired end sequencing, the larger the proportion, the greater the proportion of improperly paired reads with LL / RR pair orientation, and the smaller the proportion with RL pair orientation. Range: 0 to 1.
meanLogSRCRLen	Mean of log scaled length of the short reverse complementary regions (SRCR) in artifact chimeric fragments. SRCRs links two ss-DNA together, yielding artifact chimeric fragments. The length of SRCR follows a log-normal distribution. See rlnorm for more details. Unit: base pair (bp).
sdLogSRCRLen	Standard deviation of log scaled length of the short reverse complementary regions.
maxSRCRLen	Maximum length of the short reverse complementary regions. Unit: base pair (bp).
meanLogSRCRDist	Mean of log scaled original genomic distance of the short reverse complementary regions(SRCR) in artifact chimeric fragments. SRCRs links two ss-DNA

	together, yielding artifact chimeric fragments. The distance of SRCR is the original genomic distance between the two short reverse complementary segments, which follows a log-normal distribution in simulation. For log-normal distribution, see rlnorm for more details. Unit: base pair (bp).
sdLogSRCRDist	Standard deviation of log scaled original genomic distance of the short reverse complementary regions(SRCR) in artifact chimeric fragments.
distWinLen	The window length used in distant ss-DNA simulation. To simulate distant ss-DNA combinations, the short reverse complementary regions(SRCR) are searched between different windows. Unit: base pair (bp).
spikeWidth	The width of chimeric read spike used to simulate distant ss-DNA combinations. In real FFPE samples, the chimeric reads formed by distant DNA combination are unevenly distributed along the chromosome. Some regions are enriched in these reads while some others are scarce. The length of these regions are of similar scale; therefore, a defined width is used for simulation. Suggested range: 1500-2000. Unit: base pair (bp).
betaShape1	Shape parameter a of beta distribution used to model the unevenly distributed distant ss-DNA combinations. The number of seeds in each "spike" follows a "U" shaped beta distribution. Use this parameter to adjust the shape of the curve. See rbeta for more details. Range: 0-1.
betaShape2	Shape parameter b of beta distribution used to model the unevenly distributed distant ss-DNA combinations. The number of seeds in each "spike" follows a "U" shaped beta distribution. Use this parameter to adjust the shape of the curve. See rbeta for more details. Range: 0-1.
sameTarRegionProb	Probability of two distant ss-DNA combination events coming from the same two different windows.
adjFactor	Increase this value if the number of simulated adjacent chimeric reads is smaller than expected ($\text{sameChrProp} * \text{adjChimProp}$), decrease if the opposite is true.
distFactor	Increase this value if the number of simulated distant chimeric reads is smaller than expected, decrease if the opposite is true.
chimMutRate	Mutation rate for each base in chimeric fragments. In the chimeric fragment formation process, biological-level errors might occur and lead to mutations on these artifact fragments. For all four basic types of nucleotides, the substitution probability is set equal. Range: 0-0.75.
noiseRate	Noise rate for each base in reads. This is used for sequencing-level errors. The probability is set equal for all four basic types of nucleotides. Range: 0-0.75.
highNoiseRate	A second noise rate for each base in reads. In some real sequencing data, some reads are much more noisy than others. This parameter can be used for this situation. Range: 0-0.75.
highNoiseProp	Proportion of reads to be simulated with highNoiseRate other than noiseRate. Range: 0-1.
pairedEnd	Simulate paired end sequencing when set to true.
prefix	Prefix for read names. When reads from different runs of simulation have to be merged, please make sure that they have different prefixes.

threads	Number of threads used. Multi-threading can speed up the process.
adjChimeric	Generate reads from adjacent ss-DNA combinations if it is set to true. If it is set to false, skip this process.
distChimeric	Generate reads from distant ss-DNA combinations if it is set to true. If it is set to false, skip this process.
normalReads	Generate reads from normal fragments if it is set to true. If it is set to false, skip this process.
overWrite	Overwrite the file if file with the same output path exists and it is set to true. If file with same output path exists and it is set to false, reads will be appended to the existing file.

Details

The NGS (Next-Generation Sequencing) reads from FFPE (Formalin-Fixed Paraffin-Embedded) samples contain numerous artifact chimeric reads (ACRs), which can lead to false positive structural variant calls. These ACRs are derived from the combination of two single-stranded DNA (ss-DNA) fragments with short reverse complementary regions (SRCRs). This function simulates these artifact chimeric reads as well as normal reads for FFPE samples within defined regions. To simplify the simulation, the genome is divided into small windows, and SRCRs are found within the same window (adjacent ss-DNA combination) or between different windows (distant ss-DNA simulation). For adjacent ss-DNA combination events, the original genomic distance between and strands of two combined SRCRs are also simulated based on real data. In the output fastq file, reads are distinguished by prefixes "adjChimeric", "distChimeric" and "Normal" in their names. The parameter PhredScoreProfile can be calculated by the function [calcPhredScoreProfile](#). To simulate whole genome sequencing (WGS) or to simulate reads on several large regions / full chromosomes, please use the function [readSimFFPE](#).

Value

NULL. Reads will be written to the output FASTQ file.

Note

When fine-tuning is needed, simulate reads from part of the regions instead of all the target regions to save the runtime. Please check the package vignette for the guidance of fine-tuning.

Author(s)

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See Also

[SimFFPE](#), [calcPhredScoreProfile](#), [readSimFFPE](#)

Examples

```
PhredScoreProfilePath <- system.file("extdata", "PhredScoreProfile1.txt",
                                     package = "SimFFPE")
PhredScoreProfile <- as.matrix(read.table(PhredScoreProfilePath, skip = 1))
```

```
colnames(PhredScoreProfile) <-  
  strsplit(readLines(PhredScoreProfilePath)[1], "\t")[[1]]  
referencePath <- system.file("extdata", "example.fasta", package = "SimFFPE")  
  
regionPath <- system.file("extdata", "regionsSim.txt", package = "SimFFPE")  
targetRegions <- read.table(regionPath)  
  
outFile <- paste0(tempdir(), "/sim3")  
targetReadSimFFPE(referencePath, PhredScoreProfile, targetRegions, outFile,  
  coverage = 80, readLen = 100, meanInsertLen=180,  
  sdInsertLen=50, enzymeCut = FALSE)
```

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